

# Sheet1

	PCR 20 µL reaction (units : uL)	Final Concentration			Primers:	
10x thermopol r	2.5	1X			f_IDT_squ_LS	
10 mM dNTPs	0.5	200 µM			r_IDT_cir_LS	
10 µM Forward	0.5	0.2 µM				
10 µM Reverse	0.5	0.2 µM				
Template (colon	0	5 ng			DNA Template:	
DMSO (optional	0	3 %			Colonies	
Taq polymerase	0.125	1.0 units/50 µl PCR				
Nuclease-free	20.875					
Total volume	25					
Number of react	6				CYC --> 5 colon	Colonies 8 to 12
					1 negative control	
WORK ON ICE						
Mastermix				Tubes	C8	
10x Thermopol	15				C9	
10 mM dNTPs	3				C10	
10 µM Forward	3				C11	
10 µM Reverse	3				C12	
DMSO (optional	0				C -	
Taq DNA Polym	0.75					
Nuclease-free	125.25					
Distribute 25 µl in tubes						
Program	Y-COL-D					
PCR program A						
Initial Denaturati	95°	7 min				
	95°	30 sec				
	52°	1 min				
35 Cycles	67°	1 min 20				
Final Extension	72°	5 min				
Hold	4°					